REMARKS

Claims 24, 25, 28, 29, 32, 33, 36, 37, 40, 41, 43, 44, 47-52, 54 and 55 are pending in the present application. Claims 24, 25, 28, 29, 32, 33, 36 and 37 are withdrawn by the Examiner as being directed to different inventions. Claims 51 and 52 are canceled herein without prejudice or disclaimer. Claims 40, 41 and 47-52 are amended herein for clarity to more particularly define the invention. New claims 57-64 are added herein. In addition, applicants have made voluntary amendments to the specification on pages 13, 15, and 16 to correct inadvertent typographical errors and formatting errors. Support for these amendments and new claims can be found throughout the specification and in the language of the original claims, at least, for example, in claims 16-18, and in Figures 12, 17 and 18.

It is believed that no new matter is added by these amendments and new claims and their entry and consideration are respectfully requested. In light of these amendments and new claims and the following remarks, applicant respectfully requests reconsideration of this application and allowance of the pending claims to issue.

I. Recordation of Interview Summary

To record the Interview Summary mailed on November 25, 2009 regarding the above-referenced patent application, applicants concur that the Interview Summary accurately reflects the substance of the telephone interview that took place on November 19, 2009, in which Examiner Amanda Marie Shaw, Supervisory Examiner Dave T. Nguyen, and applicant's representatives, Dr. Alice M. Bonnen and Dr. Robert A. Schwartzman, participated.

II. Rejection under 35 U.S.C. § 102(a) and 102(e).

The Office Action states that claims 40, 44, 47, and 54 are rejected under 35 U.S.C. §102 as allegedly anticipated by Beckman et al. (U.S. Patent Application Publication No. 2003/0134307). Specifically, the Office Action states that Beckman et al. teaches a molecular beacon (MB) probe comprising standard deoxyribonucleotides and one or more 2'-O-methyl nucleotides at its 5' end. The Office Action further states that because the 5' end of the MB probe would be part of the stem region, then Beckman et al. exemplifies a MB probe comprising a stem comprising one 2'-O-methyl nucleotide and one or more unmodified nucleotides.

Applicants respectfully disagree with this interpretation of Beckman et al. However, in order to expedite the prosecution of this application, claim 40 as presented herein recites a molecular beacon probe, comprising a stem comprising one or more unmodified nucleotides, and in the 3' strand of said stem, one or more nucleotides or nucleotide analogues having an affinity increasing modification, wherein said one or more nucleotides or nucleotide analogues are selected from the group consisting of a 2'-O-derivatized nucleotide, a locked nucleic acid, and a peptide nucleic acid, wherein each base pair of said stem comprises no more than one 2'-O-derivatized nucleotide, and further wherein said probe has better stability and does not open spontaneously in the presence of contaminants present in an amplification enzyme mixture comprising said molecular beacon probe as compared to a molecular beacon probe without said stem. Support for this amendment can be found throughout the specification, at least, for example, in Figure 12.

Thus, as presented herein, claim 40 recites that the modified nucleotides are in the 3' strand of the stem of the MB probe. Beckman et al. fails to teach a MB probe having modified nucleotides only in the 3' strand of the stem, therefore, the claimed invention cannot be anticipated by Beckman et al. Further, during the interview held on November 19, 2009, it was indicated by Examiner Shaw and Examiner Nyugen that such an amendment may be sufficient to overcome this rejection. Accordingly, applicants submit that this rejection is overcome and respectfully request its withdrawal.

III. Rejection under 35 U.S.C. § 103.

The Office Action states that claims 41, 43, 48-52, and 55 are rejected under 35 U.S.C. §103(a) as allegedly unpatentable over Beckman et al. in view of Majlessi et al. (*Nucleic Acids Res.* 25:2224-2229 (1998)) and Tourkas et al. (*Nucleic Acids Res.* 30:5168-5174 (2002)). The Office Action states that Beckman et al. teaches that a MB probe can comprise one or more 2'-O-methyl nucleotides (e.g., at its 5' end). The Office Action then provides a lengthy list of what Beckman et al. does not teach and concludes that designing probes which are equivalents to those being claimed is considered "routine experimentation especially since MB probes comprising standard deoxyribonucleotides and one or more 2'-O-methyl nucleotides had already been described by Beckman" and the advantages of the 2'-

O-methyl nucleotides were known. The Office Action cites Majlessi et al. and Tourkas et al. for the advantages of 2'-O-methyl nucleotide probes over 2' deoxynucleotide probes. Office Action, pages 5-6.

As presented herein, claim 41 recites a molecular beacon probe comprising a stem and a loop, wherein said loop comprises one or more nucleotides and/or nucleotide analogues that have an affinity increasing modification, and one or more unmodified nucleotides; and said stem comprises one or more unmodified nucleotides, and in the 3' strand of said stem, one or more 2'-O-methyl nucleotides, wherein each base pair of said stem comprises no more than one 2'-O-methyl nucleotide, wherein the sensitivity of said probe to polymorphisms in the target nucleic acid sequence is lowered as compared to a molecular beacon probe without said loop and wherein the spontaneous opening of the probe in the presence of contaminants present in an amplification enzyme mixture comprising said molecular beacon probe is lowered as compared to a molecular beacon probe without said stem. Support for the amendment can be found throughout the specification, at least, for example, in Figure 12. Dependent claim 50 recites that only one base pair of the stem of the molecular beacon comprises no nucleotide or nucleotide analogue having an affinity increasing modification.

As discussed above, Beckman et al. fails to teach or suggest a MB probe having modified nucleotides in the 3' strand of the stem of the probe as claimed in the invention. Beckman et al. also fails to teach or suggest a MB probe comprising a stem wherein one base pair of the stem comprises no nucleotide or nucleotide analogue having an affinity increasing modification as claimed in the present invention. Further, during the interview held on November 19, 2009, it was indicated by Examiner Shaw and Examiner Nyugen that either amendment may be sufficient to overcome this rejection.

Majlessi et al. and Tsourkas et al. fail to remedy the deficiencies of Beckman et al. Majlessi et al. does not teach MB probes but rather teaches linear probes consisting entirely of 2'-O-methyl nucleotides and Tsourkas et al. teaches MB probes consisting entirely of 2'-O-methyl nucleotides. Majlessi et al. and Tsourkas et al. compare their probes with probes consisting only of 2'-O-deoxy nucleotides (linear and MB, respectively) and find that there are advantages of using probes consisting entirely of 2'-O-methyl nucleotides over those consisting entirely of 2'-deoxynucleotides. Majlessi et

al. discusses the advantages of the linear probes only in relationship to the binding of the probe to the target DNA/RNA. Thus, the higher T_ms, affinities, and hybridization kinetics are related only to hybridization of the probes to the target nucleic acid and not to hybridization between two strands (5' and 3' ends) of the probe itself. Thus, Majlessi et al., which relates to linear probes consisting entirely of 2'-O-methyl nucleotides and their binding to target nucleic acids, fails to provide one of ordinary skill in the art any motivation to produce the MB probes of the present invention which comprise a loop having modified and unmodified nucleotides and a stem comprising unmodified nucleotides and in the 3' strand of the stem, one or more modified nucleotides.

Similar to Majlessi et al., Tsourkas et al. also discusses the advantages of probes comprised entirely of 2'-O-methyl nucleotides as compared with probes comprised entirely of 2'-deoxynucleotides and the ability of the 2'-O-methyl nucleotide MB probes to hybridize with higher affinity and faster kinetic hybridization rates to the target nucleic acid as compared with 2'-deoxynucleotide MB probes. Tsourkas et al. also discloses that the stem-loop structure of a 2'-O-methyl nucleotide MB probe is more stable than that of MB probes comprised entirely of 2'-deoxynucleotides (page 5169, first full paragraph and page 5170, second column, first full paragraph). This greater stability of the stem-loop structure is taught by Tsourkas et al. to be the result of the 2'-O-methyl/2'-O-methyl interactions (Tsourkas et al., page 5173, first column, last sentence). Thus, in order to achieve a stable MB probe as taught by Tsoukas et al., at least the stem of the MB probe must consist entirely of 2'-O-methyl nucleotides so that when the stem is hybridized to itself (forming the hairpin structure), the 2'-O-methyl nucleotides are able to hybridize with other 2'-O-methyl nucleotides. Thus, Tsourkas et al., fails to provide one of ordinary skill in the art any motivation to produce the MB probe of the present invention, which comprises a loop comprising modified and unmodified nucleotides, and a stem comprising unmodified nucleotides and at the 3' strand of the stem, one or more modified nucleotides. In fact, by teaching that the stability of the MB probe is the result of the 2'-O-methyl/2'-O-methyl interactions, Tsourkas et al. teaches away from the present invention of having 2'-O-methyl nucleotides hybridized only to unmodified nucleotides and/or at least one base pair of the stem comprising no nucleotides or nucleotide analogues having an affinity increasing modification.

Such a teaching away in the art must be given appropriate consideration as evidence of non-obviousness, as specifically set forth in the Examination Guidelines, which state on page 57528, third column, that "[w]hen the prior art teaches away from combining certain known elements, discovery of successful means of combining them is more likely to be nonobvious.[48]" Footnote 48 as presented in the Examination Guidelines states as follows.

> [48]*United States v. Adams*, 383 U.S. 39, 51-52, 148 USPQ 479, 483 (1966). In *Adams*, the claimed invention was a battery with one magnesium electrode and one cuprous chloride electrode that could be stored dry and activated by the addition of plain water or salt water. Although magnesium and cuprous chloride were individually known battery components, the Court concluded that the claimed battery was nonobvious. The Court stated that "[d]espite the fact that each of the elements of the Adams battery was well known in the prior art, to combine them as did Adams required that a person reasonably skilled in the prior art must ignore" the teaching away of the prior art that such batteries were impractical and that water-activated batteries were successful only when combined with electrolytes detrimental to the use of magnesium electrodes. *Id.* at 42-43, 50-52, 248 USPQ at 480-483.

In like analysis of the present case, to combine the elements of the present invention, i.e., 1) 2'-O-methyl nucleotides hybridized only to unmodified nucleotides and, 2) in some embodiments, at least one base pair of the stem comprising no nucleotides or nucleotide analogues having an affinity increasing modification, would require that a person reasonably skilled in the art ignore the teaching away of Tsourkas et al. that "the stability of the MB probe is the result of the 2'-O-methyl/2'-O-methyl interactions."

Thus, this teaching away, in combination with the lack of disclosure in Beckman, Majlessi et al. or Tsourkas et al. of all of the recited elements set forth in the invention claimed herein, demonstrates that one of ordinary skill in the art would not have been motivated to even try to produce the claimed invention.

According to the interpretation of Beckman et al. as presented in the Office Action, the modified nucleotides of the Beckman et al. MB probe would be in the 5' end of the probe and would be hybridized to a non-modified nucleotide at the 3' end of the probe when the probe is in its stem-loop

(or quenched) form. The Examiner concedes that such a structure would fail to block the nuclease activity of Taq polymerase, the most commonly used enzyme in polymerase chain reactions (PCR), but reasons that such a structure would block "true" exonucleases, which cut at the 5' end of a nucleic acid strand in a sequential manner. Tsourkas et al. teaches that nuclease activity is reduced by synthesizing MBs with nuclease resistant backbones composed entirely of 2'-O-methyl modifications (page 5168, second column). Tsourkas et al. teaches that MBs consisting entirely of 2'-O-methyl modified nucleotides resist nuclease degradation and do not form a substrate for RNAse H. Thus, one of ordinary skill in the art would not be motivated to provide the MB probes as claimed in the present invention, which comprise a loop having modified and unmodified nucleotides and a stem comprising unmodified nucleotides and in the 3' strand of the stem, one or more modified nucleotides and wherein the stem can further comprise at least one base pair having no modified nucleotides since according to the cited art such probes would be less stable and more sensitive to attack from a wide variety of nucleases.

Further, none of the cited references provide any reasonable expectation of success in constructing an MB probe as claimed in the present invention. Contrary to the teachings of the cited references, applicants have unexpectedly discovered that the designing of a MB probe, which has better stability and which does not open spontaneously, depends both on the presence and position of the nucleotide analogues in the stem and whether the nucleotide analogues are base-paired with other nucleotide analogues or with unmodified nucleotides. See, for example, Table 6 of Example 4 of the present specification, which shows that the use of MB probes consisting entirely of base pairs having only one type of nucleotide (unmodified or 2'-O-methyl nucleotides) results in high levels of spontaneous opening of the probe. Notably, the MB4 probe having all modified nucleotides has a greater percentage of spontaneous opening (IBL-Increase of Baseline) than Reference MB, which is comprised entirely of unmodified nucleotides. The MB4 probe also has a greater percentage of spontaneous opening as compared to MB probes comprising a combination of unmodified and modified nucleotides. MB probes having 2'-O-methyl nucleotides base-paired with unmodified nucleotides also show increased stability, which is surprising in view of what was known in the art at the time the present invention was made. *See* Tsourkas et al., page 5173, first column, last sentence

(teaching that the greater stability of the stem-loop structure of the MB probes is the result of the 2'-O-methyl/2'-O-methyl interactions). Furthermore, as demonstrated with probes MB8 and MB9 (Figures 17 and 18, respectively), having one base pair in the stem of the MB that is comprised of unmodified nucleotides, results in an unexpectedly low level of spontaneous opening as compared with probes not having such structure (see, Example 4, Table 6, and Figures 17 and 18). Such a result could not have been predicted based on the teachings or suggestions of the cited references or based on what was commonly known at the time the present application was filed.

Thus, none of the cited art teaches or suggests that the content and placement of the modified nucleotides in a MB probe with respect to unmodified nucleotides would play a role in the functional features of a MB probe. Due to their lack of teachings, none of the cited references provide one of ordinary skill in the art any motivation to provide the MB probes of the present invention or any reasonable expectation of success in achieving the presently claimed invention.

Accordingly, in view of the foregoing, applicants respectfully submit that Beckman et al., Majlessi et al. and Tourkas et al., alone or in combination, fail to teach or suggest the presently claimed invention, and thus, request the withdrawal of this rejection.

IV. New Claims

New claims 57-64 are added herein. Support for these claims can be found in the language of the original claims and throughout the specification, at least, for example, in original claims 11, 12, 15, and 17 and in Figures 17 and 18. Thus, no new matter is believed to be added by these new claims. Further, claims 57-64 are believed to be free of the pending rejections for the same reasons set forth above explaining why claims 40, 41, 43, 44, 47-52, 54 and 55 are free of the pending rejections. Accordingly, the entry and allowance of these new claims is respectfully requested.

V. Rejoinder of method claims

Applicants request that method claims 24, 25, 28, 29, 32, 33, 36 and 37 be rejoined and examined in the present application, once a determination is made that the composition claims under

examination are allowable. Specifically, if composition claims 40, 41, 43, 44, 47-52, 54 and/or 55 are found to be allowable, applicants request that the Examiner review and examine claims 24, 25, 28, 29, 32, 33, 36, and/or 37, which recite all of the limitations of the composition claims, according to the practice of rejoinder as set forth in Section 821.04 of the MPEP. In particular, it is stated therein that if a product claim is elected in a restriction and then found allowable, withdrawn process claims that depend from or otherwise include all the limitations of the allowable product claim are to be rejoined.

The points and concerns raised in the Action having been addressed in full herein, it is respectfully submitted that this application is in condition for allowance, which action is respectfully requested. Should there be any remaining concerns, the Examiner is encouraged to contact the undersigned attorney by telephone to expedite the prosecution of this application.

No fee is believed due. However, the Commissioner is hereby authorized to charge any deficiency or credit any overpayment to Deposit Account No. 50-0220.

Respectfully submitted,

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